

Effects of Dietary Zinc Bearing Palygorskite Supplementation on the Carcass Traits, Chemical Composition of Muscle, and Muscular Lead and Chromium Contents of Broilers

Weili Yang, Yueping Chen, Yefei Cheng, Xiaohan Li, Chao Wen and Yanmin Zhou

College of Animal Science & Technology, Nanjing Agricultural University, Nanjing, Jiangsu, P.R. China, 210095

The present study was conducted to investigate the effects of zinc (Zn) bearing palygorskite (ZnPal) inclusion on the carcass traits, chemical composition of muscle, and muscular lead (Pb) and chromium (Cr) contents of broilers. A total of 240 1-day-old Arbor Acres broiler chicks were randomly divided into 5 dietary treatments with 6 replicates of 8 chicks each. Broilers in the 5 treatments were fed a basal diet supplemented with 0 (Control group), 20, 40, 60, and 80 mg/kg Zn in the form of ZnPal for 42 days, respectively. There were no differences in the carcass yield, abdominal fat yield, subcutaneous fat thickness, and intramuscular fat width among treatments ($P > 0.05$). Compared with the control group, the eviscerated yield ($P = 0.010$) and thigh muscle yield ($P = 0.046$) were quadratically increased by the supplementation of ZnPal ($P < 0.05$). Similarly, the breast muscle yield was linearly ($P = 0.024$) and quadratically ($P = 0.011$) increased by ZnPal inclusion. The addition of ZnPal to diets of broilers also linearly ($P = 0.002$) increased fat content in the thigh. Moreover, the supplementation of ZnPal linearly and quadratically reduced the content of muscular Pb and the content of Cr in the thigh muscle ($P < 0.05$). It was concluded that ZnPal inclusion could improve carcass traits, increase fat content in the thigh, and reduce the accumulations of Pb and Cr in the muscles, and this effect was more pronounced when extra Zn dosage in the form of ZnPal was 40 mg/kg.

Key words: broilers, carcass traits, chemical composition, heavy metal, zinc bearing palygorskite

J. Poult. Sci., 54: 34–40, 2017

Introduction

Zinc (Zn) is an essential trace mineral and acts both structurally and catalytically in various metalloenzymes (Vallee and Falchuk, 1993; Gaither and Eide, 2001; Park *et al.*, 2004; Salim *et al.*, 2008). These enzymes play multiple roles in metabolism (Underwood and Suttle, 1999) and other functions, such as immune response (Kidd *et al.*, 1996) and antioxidant status maintenance (Powell, 2000). However, lots of feedstuffs are marginally deficient in Zn. Therefore, Zn is commonly supplemented to poultry diet in the form of inorganic Zn sources as Zn sulfate or Zn oxide (Sandoval *et al.*, 1997; Batal *et al.*, 2001; Huang *et al.*, 2009). Although the NRC (1994) recommends that broiler diets contain 40 mg/kg of Zn, these diets are often formulated to contain dietary Zn at about 120 mg/kg since a higher concentration of dietary Zn can reduce the possibility of Zn deficiency under commercial conditions (Burrell *et al.*, 2004). In broilers,

extra Zn supplementation, irrespective of sources, has been proved to improve carcass traits and chemical composition of muscles (Liu *et al.*, 2011).

Palygorskite (Pal) is a natural hydrated magnesium aluminum silicate clay mineral with chain-layered crystal structure, and it has considerable micropores and channels that are comprised by silicon oxygen tetrahedron and aluminium-oxygen octahedral chains (Galan *et al.*, 1994; Bergaya *et al.*, 2006; Brigatti *et al.*, 2006; Bergaya and Lagaly, 2013). The chemical and physical characteristics of Pal endow it with adsorption property, good adhesive ability and cation exchange capacity (Galan, 1996; Galan and Carretero, 1999; Murray, 2000; Carretero, 2002; Chen and Wang, 2007; Huang *et al.*, 2007; Zhou, 2011). In animal nutrition, Pal has been widely used as pellet binder, feed ingredients or feed supplement to promote growth, maintain health, enhance immunity, and detoxify toxins (Schell *et al.*, 1993; Zaid *et al.*, 1995; Pappas *et al.*, 2010; Ministry of Agriculture of China, 2013; Zhang *et al.*, 2013). Due to their potential neurotoxic, teratogenic and lethal effect, the accumulations of heavy metals including lead (Pb), chromium (Cr) and cadmium (Cd) would pose a threat to all forms of life including human, animal, plant and aquatic life through the

Received: April 8, 2016, Accepted: June 26, 2016

Released Online Advance Publication: August 25, 2016

Correspondence: Yanmin Zhou, College of Animal Science and Technology, Nanjing Agricultural University, No. 1 Weigang, Nanjing, Jiangsu 210095, P.R. China. (E-mail: zhouym6308@163.com)

food chain (Carpenter, 1987; Goyer, 1996; Ruff *et al.*, 1996; Zhuang *et al.*, 2009). Previous studies have shown that Pal, as an effective solution, could be used for the removal of heavy metals both *in vitro* (Alvarez-Ayuso and Garcia-Sanchez, 2003; Potgieter *et al.*, 2006; Fan *et al.*, 2009) and *in vivo* (Zhang *et al.*, 2015; Cheng *et al.*, 2016).

Zn bearing Pal (ZnPal) has been recently prepared using solid state ion exchange method (Yan *et al.*, 2016a, 2016b). In an *in vivo* study, Yan *et al.* (2016a) have newly found that ZnPal as a potential Zn source displayed a higher bio-availability than Zn sulfate during a 42-day study in broilers. In a previous study (Yang *et al.*, 2016b), it has been demonstrated that ZnPal supplementation could also improve meat quality and its antioxidant capacity whereas did not affect growth performance and muscular Zn content of broilers. However, little was known about extra ZnPal supplementation on carcass and meat composition, and the residues of heavy metals in muscles of broilers. Therefore, the current study was conducted to investigate the effects of ZnPal inclusion on carcass traits, chemical component of muscle, and the accumulations of muscular Pb and Cr in broilers.

Materials and Methods

Preparation of ZnPal

The Pal was kindly provided by Jiangsu Sinitic Biotech Co., Ltd. (Xuyi, Jiangsu, P. R. China) and sieved by a 200-mesh sieve (diameter, 0.074 mm). The main chemical compositions of Pal determined by a Minipal 4X-ray fluorescence spectrometer (PANalytical, Netherland) are listed in the following: SiO₂, 59.11%; MgO, 12.75%; Al₂O₃, 10.31%; CaO, 7.42%; Fe₂O₃, 6.21%; Na₂O, 1.27% and K₂O, 1.19%.

ZnPal was prepared using solid state ion exchange method as previously described by Yan *et al.* (2016). In detail, Pal was firstly calcinated at around 300°C for 1 h in the muffle oven. After cooling down, Pal was mixed with ZnCl₂ (ZnCl₂ ≥ 98.0%; 4:1, wt/wt) purchased from Nanjing Chemical Reagent Co., Ltd. (Nanjing, Jiangsu, P. R. China) in a stainless steel blade grinder. The mixture was subsequently calcinated at 300°C for 3 h in a muffle oven. After cooling down to room temperature, the mixture was washed repeatedly by deionized water until there was no white deposition generated in the washed solution when swigged with

Table 1. Composition and nutrient level of basal diet (g/kg, as fed basis unless otherwise stated)

Items	1-21 days	22-42 days
Ingredients		
Corn	576.1	622.7
Soybean meal	310	230
Corn gluten meal	32.9	60
Soybean oil	31.1	40
Limestone	12	14
Dicalcium phosphate	20	16
L-Lysine	3.4	3.5
DL-Methionine	1.5	0.8
Sodium chloride	3	3
Premix ^a	10	10
Calculated nutrient levels		
Apparent metabolizable energy (MJ/kg)	12.56	13.19
Crude protein	211	196
Calcium	10.00	9.50
Available phosphorus	4.60	3.90
Lysine	12.00	10.50
Methionine	5.00	4.20
Methionine + cystine	8.50	7.60
Analyzed composition ^b		
Crude protein	208	192
Ash	57.2	56.5

^a Premix provided per kilogram of diet: vitamin A (transretinyl acetate), 10,000 IU; vitamin D₃ (cholecalciferol), 3,000 IU; vitamin E (all-rac- α -tocopherol), 30 IU; menadione, 1.3 mg; thiamin, 2.2 mg; riboflavin, 8 mg; nicotinamide, 40 mg; choline chloride, 400 mg; calcium pantothenate, 10 mg; pyridoxine·HCl, 4 mg; biotin, 0.04 mg; folic acid, 1 mg; vitamin B₁₂ (cobalamin), 0.013 mg; Fe (from ferrous sulfate), 80 mg; Cu (from copper sulphate), 8.0 mg; Mn (from manganese sulphate), 110 mg; Zn (from zinc oxide), 60 mg; I (from calcium iodate), 1.1 mg; Se (from sodium selenite), 0.3 mg.

^b Values based on analysis of triplicate samples of diets.

Table 2. Analyzed mineral elements content in the diets (mg/kg)

Items ^{1,2}	Zn as ZnPal (mg/kg)				
	0 (Basal diet)	20	40	60	80
1-21 d					
Zn	81±2	102±3	120±2	144±4	165±3
Fe	277±4	280±7	276±5	283±7	286±6
Cu	22.8±0.3	21.9±0.7	22.5±0.5	22.9±0.6	23.5±0.4
Mn	164±6	163±3	168±4	170±5	174±6
Mg	1825±13	1821±19	1848±14	1831±12	1817±14
Pb	2.15±0.19	2.05±0.09	2.17±0.28	1.98±0.08	2.13±0.06
Cr	1.26±0.18	1.01±0.21	0.96±0.08	1.08±0.13	1.20±0.03
22-42 d					
Zn	88±4	106±5	128±3	146±3	170±2
Fe	271±6	264±5	260±4	274±7	283±4
Cu	24.8±1.0	23.5±0.8	22.7±0.6	23.9±0.4	23.0±0.5
Mn	194±6	197±4	186±4	188±5	193±7
Mg	1819±17	1835±11	1825±18	1856±22	1885±19
Pb	1.93±0.12	2.01±0.06	1.83±0.11	1.79±0.08	1.91±0.17
Cr	1.21±0.09	1.24±0.19	1.17±0.14	1.31±0.11	1.19±0.07

¹ Values based on analysis of triplicate samples of diets.

² Means and standard error were presented.

0.1 mol/L AgNO₃ solution. Finally, the washed mixture were collected and dried at around 105°C for 2 h in an air oven, and then ground through a 200-mesh sieve after cooling down. The amount of Zn adsorbed by Pal was 47.15 mg/g.

Experimental Design, Diets and Management

All procedures were approved by Nanjing Agricultural University Institutional Animal Care and Use Committee. A total of 240 1-day-old Arbor Acres broiler chicks (initial weight, 36.71±0.18 g) obtained from a commercial hatchery were randomly allocated into 5 dietary treatments with 6 replicates (cages) of 8 chicks each (4 males and 4 females/cage). Birds in the 5 treatments were given a basal diet supplemented with 0 (control group), 20, 40, 60, and 80 mg/kg Zn diet as ZnPal for 42 days, respectively. The formulation and calculated nutrient level of basal diet are shown in Table 1. The measured mineral elements contents in the diets are shown in the Table 2. Birds were allowed *ad libitum* access to mash feed and water. The water met the national drinking water standard of China (Ministry of Health of China, 2006), in which the concentrations of As <0.01 mg/L, Cd<0.005 mg/L, Cr (VI)<0.05 mg/L, Pb<0.01 mg/L, Hg<0.001 mg/L, Zn<1.0 mg/L. All birds were placed in wire cages and housed in an environmentally controlled room with continuing light. Temperature was maintained at 32 to 34°C for the first 3 days and then reduced by 2 to 3°C per week to a final temperature of 20°C.

Carcass Traits

At 42 days of age, 1 male bird from each replicate (cage) was randomly selected and weighed after feed deprivation for 12 h. Chickens were euthanized by cervical dislocation. Subcutaneous fat thickness and intramuscular fat width were measured by a vernier caliper as previously described (Wu *et al.*, 2012). The hot carcasses were weighted after bleeding

and defeathering. The head, feet, abdominal fat (fat surrounding the cloaca and the gizzard), and all of the viscera except the kidney were then further removed to determine eviscerated yield based on live weight. The left pectoralis major muscle and thigh muscle were excised without skin and immediately weighed to calculate the yield based on eviscerated weight. After that, the left pectoralis major muscle and thigh muscle samples were immediately frozen and stored at -20°C for further analysis.

Chemical Composition of Muscles

The contents of moisture, crude protein and crude fat of muscle samples (pectoralis major muscle and thigh muscle) were determined according to the standardized procedures as described by Zhang (2003).

Determination of Muscular Mineral Elements

The contents of Zn, Pb, Cr, Cd, and mercury (Hg) in the muscle samples (left major muscle and thigh muscle) were determined according to the method described by Tang *et al.* (2015) and Yan *et al.* (2016a). Approximately 2.0 g of muscle sample was firstly weighed into a glass digestion tube, mixed with 10 mL of a mixture acid of nitric acid and perchloric acid (4:1, vol/vol) at room temperature for 12 h, and then digested on a heating block (LabTech DigiBlock Digester, EHD36, Labtech Co., Ltd., P. R. China) to acquire a clear digested solution. The procedure of digestion was in the following: 90°C for 30 min; 120°C for 30 min; 160°C for 120 min; 180°C for 180 min. After that, digested solutions were diluted with ultra-pure water to a final volume of 25 mL. The contents of Zn, Pb, Cr, Cd and Hg in the digested liquid were determined using ICP-MS (Optimal 2100DV, Perkin-Elmer-Sciex, Norwalk, NY, USA). The operating conditions are presented in the following: power, 1300 W; plasma gas flow rate, 12 L/min; auxiliary gas flow rate, 0.2 L/min; nebuliser gas flow rate, 0.55 L/min; sample flow rate,

Table 3. The carcass traits of broilers at 42 days of age

Items	Zn as ZnPal ¹ (mg/kg)					SEM ²	P-value		
	0 (Control)	20	40	60	80		Control vs ZnPal	Linear	Quadratic
Carcass yield (g/kg)	928.2	930.9	927.6	925.5	922.1	1.5	0.428	1.440	0.229
Eviscerated yield (g/kg)	668.7 ^{ab}	674.3 ^{ab}	686.9 ^a	679.6 ^{ab}	654.7 ^b	3.2	0.034	0.898	0.010
Breast muscle yield (g/kg)	256.3 ^b	275.4 ^{ab}	301.1 ^a	295.2 ^a	276.5 ^{ab}	4.0	0.008	0.024	0.011
thigh muscle yield (g/kg)	173.3	185.6	203.9	187.9	176.1	4.2	0.210	0.532	0.046
Abdominal fat yield (g/kg)	17.63	17.30	17.35	20.79	19.31	0.91	0.707	0.347	0.697
Subcutaneous fat thickness (mm)	8.18	8.06	10.27	10.12	8.10	0.40	0.148	0.526	0.174
Intramuscular fat width (mm)	9.69	9.67	8.82	8.76	9.13	0.28	0.755	0.332	0.750

^{a-b} Means within a row with different superscripts are different at $P < 0.05$.

¹ ZnPal, zinc bearing palygorskite.

² SEM, total standard error of means.

Table 4. Muscle composition of broilers at 42 days of age (%)

Items		Zn as ZnPal ¹ (mg/kg)					SEM ²	P-value		
		0 (Control)	20	40	60	80		Control vs ZnPal	Linear	Quadratic
Moisture	Breast	72.88	72.57	72.64	72.23	72.77	0.15	0.715	0.574	0.418
	Thigh	72.48	72.54	73.27	73.37	71.82	0.33	0.628	0.986	0.299
Crude fat	Breast	1.71	1.42	1.54	1.52	1.35	0.08	0.738	0.292	0.876
	Thigh	2.52 ^b	3.28 ^{ab}	2.98 ^{ab}	3.52 ^a	3.58 ^a	0.11	0.016	0.002	0.676
Crude protein	Breast	23.18	23.95	23.55	23.87	23.65	0.18	0.735	0.440	0.429
	Thigh	21.67	22.29	21.84	21.04	22.59	0.33	0.644	0.863	0.814

^{a-b} Means within a row with different superscripts are different at $P < 0.05$.

¹ ZnPal, zinc bearing palygorskite.

² SEM, total standard error of means.

1.5 mL/min; sample uptake rate, 1.0 mL/min. Validation of the minerals analysis was conducted using a certified bovine liver powder (GBW (E) 080193; National Institute of Standards and Technology, Beijing, P. R. China) as a standard reference material. The Cd and Hg were not detected in the muscles and diets, and therefore are not presented in the Tables.

Statistical Analysis

Data were analyzed by one-way ANOVA using SPSS (2008) statistical software (Ver. 16.0 for Windows, SPSS Inc., Chicago, IL, USA). Polynomial contrasts were used to determine the linear and quadratic effects of dietary ZnPal inclusion level. The differences among treatments were examined by Tukey's test, which were considered to be significant at $P < 0.05$. The means and total standard error of means (SEM) were presented.

Results

Carcass Traits

Compared with the control group (Table 3), the eviscerated yield ($P = 0.010$) and thigh muscle yield ($P = 0.046$) were quadratically increased by the inclusion of ZnPal. Likewise, the addition of ZnPal linearly ($P = 0.024$) and quadratically ($P = 0.011$) increased breast muscle yield.

However, no differences were observed in carcass yield, abdominal fat yield, subcutaneous fat thickness, and intramuscular fat width among groups ($P > 0.05$).

Chemical Composition of Muscle

The supplementation of ZnPal linearly increased the fat content in thigh muscle (Table 4, $P = 0.002$) whereas the similar effect was not observed for the fat content in the breast muscle ($P > 0.05$). Additionally, neither muscular moisture nor crude protein was altered by ZnPal inclusion ($P > 0.05$).

Muscular Mineral Elements

As indicated in the Table 5, the supplementation of ZnPal linearly and quadratically reduced the content of muscular Pb and the content of Cr in the thigh muscle ($P < 0.05$). No difference was observed in the content of Cr in breast muscle ($P > 0.05$).

Discussion

In broilers, Saenmahayak *et al.* (2010) found that complexed Zn supplementation did not influence carcass traits and component yields whereas increased deboned fillet and total breast yields. In the current study, the growth performance of broilers at 42 days of age was similar among groups, and the live weight of broilers at 42 days of age in the

Table 5. The contents of heavy toxic metals in breast and thigh muscles (mg/kg fresh sample)

Items	Zn as ZnPal ¹ (mg/kg)					SEM ²	P-value		
	0 (Control)	20	40	60	80		Control vs ZnPal	Linear	Quadratic
Breast									
Pb	0.124 ^a	0.118 ^{ab}	0.113 ^b	0.112 ^b	0.120 ^{ab}	0.001	0.015	0.039	0.016
Cr	0.286	0.253	0.254	0.290	0.313	0.010	0.357	0.406	0.070
Thigh									
Pb	0.120 ^a	0.128 ^a	0.123 ^a	0.083 ^b	0.083 ^b	0.004	<0.001	<0.001	<0.001
Cr	0.281 ^{ab}	0.300 ^a	0.266 ^{ab}	0.242 ^{bc}	0.218 ^c	0.007	<0.001	<0.001	0.003

^{a-b} Means within a row with different superscripts are different at $P < 0.05$.

¹ ZnPal, zinc bearing palygorskite.

² SEM, total standard error of means.

control group and the four ZnPal groups (from 20 to 80 mg/kg Zn) was 2253, 2272, 2233, 2200, and 2309 g, respectively (Yang *et al.*, 2016). However, dietary ZnPal supplementation increased eviscerated yield and muscle yield, indicating that the inclusion of ZnPal could improve the carcass composition of broilers. The increased eviscerated yield resulting from Zn supplementation may be in related with the simultaneously improvement of muscle yield. This result was in agreement with finding of Liu *et al.* (2011) who reported that extra Zn supplementation, irrespective of sources, increased the percentage of eviscerated yield in broilers. Similar results were also observed by Sahin *et al.* (2005) in Japanese quail, and Spears and Kegley (2002) in steers. The improved carcass traits induced by the supplementation of ZnPal may be associated with the biological function of Zn which is involved in endocrine function such as the synthesis and secretion of various hormones including osteocalcin, testosterone, thyroid hormones, insulin-like growth factor-1, and insulin, which play vital roles in the growth and development of bone and muscle (Giugliano and Millward, 1984; Ploysangam *et al.*, 1997; Fukada *et al.*, 2011). Huang *et al.* (2007) demonstrated that the optimal dietary Zn of chicks from hatch to 21 days of age was 84 mg/kg. Similarly, Sunder *et al.* (2008) reported that basal diet Zn (29 ppm) was adequate to support optimum performance of broilers, and 80 ppm of Zn was required up to 4 wk of age for better mineral retention, immune response, and alleviation of stress. These studies together showed that a higher intake of Zn that exceeded the NRC recommendation (40 mg/kg) was beneficial for the growth of broilers. In this study, the carcass characteristics of broilers were optimized by the extra Zn supplementation at the dosage of 40 mg/kg as ZnPal, which in turn suggested that a higher level of Zn intake was required for the optimal carcass composition.

Muscular fat is very important from the sensory aspect since it is a source of many aromatic substances affecting the meat taste (Suchý *et al.*, 2002). In this study, dietary ZnPal supplementation linearly increased the fat content in thigh. Likewise, Liu *et al.* (2011) found that the addition of extra Zn to broiler diet increased the dry matter and intramuscular fat contents of the breast muscle. Additionally, Greene *et al.*

(1988) observed that the addition of organic Zn (Zn-Met) increased USDA carcass quality grade, external fat, and kidney, pelvic, and heart fat of steers. Previous studies have shown that the supplementation of Zn bearing clay improved the intestinal morphology and digestibilities of nutrients (Hu *et al.*, 2013; Tang *et al.*, 2014). Thus, the improved muscle composition may be associated with the possibly enhanced nutrient availability resulting from ZnPal supplementation.

In *in vitro* studies, Pal used as an adsorbent can remove Pb and Cr in heavy metal contained water or soil (Alvarez-Ayuso and Garcia-Sanchez, 2003; Potgieter *et al.*, 2006; Fan *et al.*, 2009). In the current study, dietary ZnPal did not alter muscular Zn content (Yang *et al.*, 2016), and it may due to that muscle, unlike pancreas and tibia, was not sensitive to reflect Zn retention (Huang *et al.*, 2007, 2009). However, the supplementation of ZnPal in this study reduced the accumulations of Pb and Cr in the muscles, and it indicated that ZnPal supplementation could improve the safety of meat products. Similar results were also observed by Cheng *et al.* (2016) who reported that dietary Pal inclusion, at either 10 or 20 g/kg, significantly decreased Pb accumulation in the breast or thigh muscle. Additionally, Zhang *et al.* (2015) reported that 2% Pal supplementation reduced muscular Cd accumulation in blunt snout bream. Inclusion of diets with excess minerals could lead to antagonism. The antagonistic effects between Zn and heavy metals have been well demonstrated (Waalke and Poirier, 1984; Fosmire, 1990; Kargin and Çogun, 1999; McDonald *et al.*, 2011). Thus, the reduced accumulation of heavy metals may also result from the antagonism induced by extra Zn supplementation in the form of ZnPal.

The newly total maximum Zn content recommended by EU are 150 mg/kg Zn complete feed for piglets, sows, rabbits, salmonids, cats and dogs; 120 mg/kg Zn complete feed for turkeys for fattening; 100 mg/kg Zn complete feed for all other species and categories, and it would result in an overall reduction of Zn emission from animal production of about 20% (EFSA, 2014). Under commercial conditions, diets for broiler chickens are always designed to contain Zn at approximately 120 mg/kg to acquire the optimal growth performance (Burrell *et al.*, 2004). However, the major of

ingested Zn (around 94%) in broilers is excreted actually, and it can in turn lead to Zn resource waste, and subsequently increase Zn load on the environment (Mohanna and Nys, 1997, 1999). To avoid these problems, Zn sources with a higher bioavailability would be a new prospect. Clays (as Pal and zeolite) with plentiful pores, high aspect ratio, high specific surface area, and good ion-exchange capacity can be used as carriers of active components (Murray, 2000; Tang *et al.*, 2015; Yan *et al.*, 2016a). In broilers, Yan *et al.* (2016b) have shown that Zn-Pal had a higher bioavailability than ZnSO₄ in broiler diets, and the optimal level of supplemented Zn for broilers was 60 mg/kg in the form of Zn-Pal. Previous studies have also indicated, zeolite bearing Zn exhibited a higher bioavailability than ZnSO₄ in broilers (Tang *et al.*, 2015) and laying hens (Li *et al.*, 2015). These studies together indicated that clay bearing Zn may be a prospect Zn source in the future.

In conclusion, the present study demonstrated that Zn supplementation as ZnPal (from 0 to 80 mg/kg) especially at the dosage of 40 mg/kg improved the eviscerated and muscle yields, increased the fat content in thigh muscle, and decreased the accumulation of Pb and Cr in breast and thigh muscles.

References

- Alvarez-Ayuso E and Garcia-Sanchez A. Palygorskite as a feasible amendment to stabilize heavy metal polluted soils. *Environmental Pollution*, 125: 337–344. 2003.
- Bao YM, Choct M, Iji PA and Bruerton K. Trace mineral interactions in broiler chicken diets. *British Poultry Science*, 51: 109–117. 2010.
- Batal B, Parr TM and Baker DH. Zinc bioavailability in tetrabasic zinc chloride and the dietary zinc requirement of young chicks fed a soy concentrate diet. *Poultry Science*, 80: 87–90. 2001.
- Bergaya F and Lagaly G. *Handbook of Clay Science*, 2th Edition. Developments in Clay Science vol. 5. Elsevier, Amsterdam, 2013.
- Bergaya F, Theng BKG and Lagaly G. History of clay science: a young discipline. In: Bergaya F, Theng BKG, Lagaly G. (Eds.), *Handbook of Clay Science*, 1st ed. Elsevier, Amsterdam, The Netherlands. 2006.
- Brigatti MF, Galan E and Theng BKG. Structures and mineralogy of clay minerals. *Handbook of Clay Science*, 1: 19–69. 2006.
- Burrell AL, Dozier WA, Davis AJ, Compton MM, Freeman ME, Vendrell PF and Ward TL. Responses of broilers to dietary zinc concentrations and sources in relation to environmental implications. *British Poultry Science*, 45: 255–263. 2004.
- Carpenter SJ. Developmental analysis of cephalic axial dysraphic disorders in arsenic-treated hamster embryos. *Anatomy and Embryology*, 176: 345–365. 1987.
- Carretero MI. Clay minerals and their beneficial effects upon human health. A review. *Applied Clay Science*, 21: 155–163. 2002.
- Chen H and Wang A. Kinetic and isothermal studies of lead ion adsorption onto palygorskite clay. *Journal of Colloid and Interface Science*, 307: 309–316. 2007.
- Cheng YF, Chen YP, Li XH, Yang WL, Wen C and Zhou YM. Effects of palygorskite inclusion on the growth performance, meat quality, antioxidant ability and mineral elements contents of broilers. *Biological Trace Element Research*, 173: 194–201. 2016.
- EFSA. Scientific opinion on the potential reduction of the currently authorised maximum zinc content in complete feed. *EFSA Journal*, 12: 3668. 2014.
- Fan Q, Li Z, Zhao H, Jia Z, Xu J and Wu W. Adsorption of Pb (II) on palygorskite from aqueous solution: effects of pH, ionic strength and temperature. *Applied Clay Science*, 45: 111–116. 2009.
- Fosmire GJ. Zinc toxicity. *The American Journal of Clinical Nutrition*, 51: 225–227. 1990.
- Fukada T, Yamasaki S, Nishida K, Murakami M and Hirano T. Zinc homeostasis and signaling in health and diseases. *JBIC Journal of Biological Inorganic Chemistry*, 16: 1123–1134. 2011.
- Gaither LA and Eide DJ. Eukaryotic Zn transporters and their-regulation. *Biometals*, 14: 251–270. 2001.
- Galan E, Mesa JM and Sanchez C. Properties and applications of palygorskite clays from Ciudad Real, Central Spain. *Applied Clay Science*, 9: 293–302. 1994.
- Galan E. Properties and applications of palygorskite–sepiolite clays. *Clay Minerals*, 31: 443–453. 1996.
- Galan E and Carretero MI. A new approach to compositional limits for sepiolite and palygorskite. *Clays and Clay Minerals*, 47: 399–409. 1999.
- Giugliano R and Millward DJ. Growth and zinc homeostasis in the severely Zn-deficient rat. *British Journal of Nutrition*, 52: 545–560. 1984.
- Goyer RA. Results of lead research: prenatal exposure and neurological consequences. *Environment Health Perspectives*, 104: 1050–1054. 1996.
- Greene LW, Lunt DK, Byers FM, Chirase NK, Richmond CE, Knutson RE and Schelling, GT. Performance and carcass quality of steers supplemented with zinc oxide or zinc methionine. *Journal of Animal Science*, 66: 1818–1823. 1988.
- Hu CH, Qian ZC, Song J, Luan ZS and Zuo AY. Effects of zinc oxide-montmorillonite hybrid on growth performance, intestinal structure, and function of broiler chicken. *Poultry Science*, 92: 143–150. 2013.
- Huang J, Liu Y, Jin Q, Wang X and Yang J. Adsorption studies of a water soluble dye, Reactive Red MF-3B, using sonication-surfactant-modified attapulgite clay. *Journal of Hazardous Materials*, 143: 541–548. 2007.
- Huang YL, Lu L, Luo XG and Liu B. An optimal dietary zinc Level of broiler chicks fed a corn-soybean meal diet. *Poultry Science*, 86: 2582–2589. 2007.
- Huang YL, Lu L, Li SF, Luo XG and Liu B. Relative bioavailabilities of organic zinc sources with different chelation strengths for broilers fed a conventional corn-soybean meal diet. *Journal of Animal Science*, 87: 2038–2046. 2009.
- Kargin F and Çogun HY. Metal interactions during accumulation and elimination of zinc and cadmium in tissues of the freshwater fish *Tilapia nilotica*. *Bulletin of Environmental Contamination and Toxicology*, 63: 511–519. 1999.
- Kidd MT, Ferket PR and Qureshi MA. Zinc metabolism with special reference to its role in immunity. *World's Poultry Science Journal*, 52: 309–324. 1996.
- Li L, Li P, Chen Y, Wen C, Zhuang S and Zhou Y. Zinc-bearing zeolite clinoptilolite improves tissue zinc accumulation in laying hens by enhancing zinc transporter gene mRNA abundance. *Animal Science Journal*, 86: 782–789. 2015.
- Liu ZH, Lu L, Li SF, Zhang LY, Xi L, Zhang KY and Luo XG. Effects of supplemental zinc source and level on growth performance, carcass traits, and meat quality of broilers. *Poultry*

- Science, 90: 1782–1790. 2011.
- McDonald P, Edwards RA, Greenhalgh JFD, Morgan CA, Sinclair LA and Wilkinson RG. *Animal Nutrition*, 7th Edition. Pearson Education Limited, Edinburgh Gate, Harlow, Essex CM20 2JE, England. pp. 103–136. 2011.
- Mohanna C and Nys Y. Excess Zinc in manure of broiler chicks: Decrease in Zinc supplementation and use of phytase improve its retention in the carcasses. *Proceedings of the 11th European Symposium on Poultry Nutrition*, Faaborg, pp. 459–461. 1997.
- Mohanna C and Nys Y. Effect of dietary zinc content and sources on the growth, body zinc deposition and retention, zinc excretion and immune response in chickens. *British Poultry Science*, 40: 108–114. 1999.
- Ministry of Agriculture of China. *Feed raw materials catalogue No. 2038*. 2013.
- Ministry of Health of China. *The standard for drinking water GB5749*. 2006.
- Murray HH. Traditional and new applications for kaolin, smectite, and palygorskite: a general overview. *Applied Clay Science*, 17: 207–221. 2000.
- Pappas AC, Zoidis E, Theophilou N, Zervas G and Fegeros K. Effects of palygorskite on broiler performance feed technological characteristics and litter quality. *Applied Clay Science*, 49: 276–280. 2010.
- Park SY, Birkhold SG, Kubena LF, Nisbet DJ and Ricke SC. Review on the role of dietary zinc in poultry nutrition, immunity, and reproduction. *Biological Trace Element Research*, 101: 147–163. 2004.
- Ploysangam A, Falciglia GA and Brehm BJ. Effect of marginal zinc deficiency on human growth and development. *Journal of Tropical Pediatrics*, 43: 192–198. 1997.
- Potgieter JH, Potgieter-Vermaak SS and Kalibantonga PD. Heavy metals removal from solution by palygorskite clay. *Minerals Engineering*, 19: 463–470. 2006.
- Powell SR. The antioxidant properties of zinc. *The Journal of Nutrition*, 130: 1447S–1454S. 2000.
- Ruff HA, Markowitz ME, Bijur PE and Rosen JF. Relationships among blood lead levels, iron deficiency, and cognitive development in 2-year-old children. *Environment Health Perspectives*, 104: 180–185. 1996.
- Saenmahayak B, Bilgili SF, Hess JB and Singh M. Live and processing performance of broiler chickens fed diets supplemented with complexed zinc. *The Journal of Applied Poultry Research*, 19: 334–340. 2010.
- Sahin K, Smith MO, Onderci M, Sahin N, Gursu MF and Kucuk O. Supplementation of zinc from organic or inorganic source improves performance and antioxidant status of heat-distressed quail. *Poultry Science*, 84: 882–887. 2005.
- Salim HM, Jo C and Lee BD. Zinc in broiler feeding and nutrition. *Avian Biology Research*, 1: 5–18. 2008.
- Sandoval M, Henry PR, Ammerman CB, Miles RD and Littell RC. Relative bioavailability of supplemental inorganic zinc sources for chicks. *Journal of Animal Science*, 75: 3195–3205. 1997.
- Schell TC, Lindemann MD, Kornegay ET, Blodgett DJ and Doerr JA. Effectiveness of different types of clay for reducing the detrimental effects of aflatoxin contaminated diets on performance and serum profiles of weanling pigs. *Journal of Animal Science*, 71: 1226–1231. 1993.
- Spears JW and Kegley EB. Effect of zinc source (zinc oxide vs zinc proteinate) and level on performance, carcass characteristics, and immune response of growing and finishing steers. *Journal of Animal Science*, 80: 2747–2752. 2002.
- SPSS, SPSS 16. 0 for Windows. SPSS Inc, Chicago, IL, USA, 2008.
- Suchý P, Jelínek P, Straková E and Hucl J. Chemical composition of muscles of hybrid broiler chickens during prolonged feeding. *Czech Journal of Animal Science*, 47: 511–518. 2002.
- Sunder GS, Panda AK, Gopinath NCS, Rao SR, Raju MVLN, Reddy MR and Kumar CV. Effects of higher levels of zinc supplementation on performance, mineral availability, and immune competence in broiler chickens. *The Journal of Applied Poultry Research*, 17: 79–86. 2008.
- Tang ZG, Wen C, Li P, Wang T and Zhou YM. Effect of zinc-bearing zeolite clinoptilolite on growth performance, nutrient retention, digestive enzyme activities, and intestinal function of broiler chickens. *Biological Trace Element Research*, 158: 51–57. 2014.
- Tang ZG, Chen GY, Li LF, Wen C, Wang T and Zhou YM. Effect of zinc-bearing zeolite clinoptilolite on growth performance, zinc accumulation, and gene expression of zinc transporters in broilers. *Journal of Animal Science*, 93: 620–626. 2015.
- Underwood EJ and Suttle NF. *The mineral nutrition of livestock*. CABI Publishing, New York, USA, 1999.
- Vallee BL and Falchuk KH. The biochemical basis of Zn physiology. *Physiological Reviews*, 73: 79–1182. 1993.
- Waalkes MP and Poirier LA. In vitro cadmium-DNA interactions: cooperativity of cadmium binding and competitive antagonism by calcium, magnesium, and zinc. *Toxicology and Applied Pharmacology*, 75: 539–546. 1984.
- Wu QJ, Wang YQ, Zhou YM and Wang T. Dietary effects of natural and modified clinoptilolite supplementation on growth performance, fat deposition and carcass characteristics of broilers. *Journal of Animal and Veterinary Advances*, 11: 4263–4268. 2012.
- Yan R, Zhang L, Yang X, Wen C and Zhou YM. Bioavailability evaluation of zinc-bearing palygorskite as a zinc source for broiler chickens. *Applied Clay Science*, 119: 155–160. 2016a.
- Yang WL, Chen YP, Cheng YF, Li XH, Zhang RQ, Wen C and Zhou YM. An evaluation of zinc bearing palygorskite inclusion on the growth performance, mineral content, meat quality, and antioxidant status of broilers. *Poultry Science*, 95: 878–885. 2016b.
- Zaid MRB, Hasan M and Khan AA. Attapulgit in the treatment of acute diarrhoea: a double-blind placebo-controlled study. *Journal of Diarrhoeal Diseases Research*, 13: 44–46. 1995.
- Zhang J, Lv Y, Tang C and Wang X. Effects of dietary supplementation with palygorskite on intestinal integrity in weaned piglets. *Applied clay science*, 86: 185–189. 2013.
- Zhang LY. *Feed analysis and quality inspection technology*. China Agricultural University Press, Beijing. 2003.
- Zhang RQ, Yang X, Chen YP, Yan R, Wen C, Liu WB and Zhou YM. Effects of feed palygorskite inclusion on pelleting technological characteristics, growth performance and tissue trace elements content of blunt snout bream (*Megalobrama amblycephala*). *Applied Clay Science*, 114: 197–201. 2015.
- Zhou CH. An overview on strategies towards clay-based designer catalysts for green and sustainable catalysis. *Applied Clay Science*, 53: 97–105. 2011.
- Zhuang P, Zou H and Shu WS. Biotransfer of heavy metals along a soil-plant-insect-chicken food chain: Field study. *Journal of Environmental Sciences*, 21: 849–853. 2009.